Field Desorption Mass Spectrometry of Pesticides and Their Metabolites

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The first application of low- and high-resolution field desorption mass spectrometry (fdms) to chlorinated pesticidal compounds of the dimethano-naphthalene and methano-indene types demonstrates the potential of this new analytical technique for the determination of the molecular weight of basic pesticides. The technique proves to be even better for their metabolites. Fdms enables positive identification of these compounds. The water elimination observed in the fd mass spectra of *cis*- and *trans*-aldrindiol can be used to distinguish between these stereoisomers, which is not possible with electron impact mass spectrometry (eims).

Most of the early work on the ei mass spectra of bridged polycyclic chlorinated pesticidal compounds was initiated by Damico *et al.* in 1968 and demonstrated the principal features of the fragmentation pattern of this ionization method. The later field ionization (fi)ms investigations by the same author in 1969 (Damico, 1969) show well the enhanced molecular ion intensities obtainable with this technique. The comparison of ei- and fims is, in this respect, a link to our fdms work.

However, there is a decisive difference between ei-, fi-, and chemical ionization (ci)ms on the one hand and fdms on the other. In contrast to all other methods of ion production for mass spectrometry, the sample in fd *is not evaporated* from the commonly-used introduction system, but rather applied to the field anode from a solution or suspension using the emitter dipping technique (Beckey, 1969). Ionization and desorption of the adsorbed molecules can in this manner be performed with minimal thermal stress, which is of special importance for highly polar compounds such as metabolites carrying a number of hydroxyl or carboxyl groups (Schulten *et al.*, 1972).

There are three main advantages of fdms in the field of metabolic investigations. (1) There is small sample consumption (in the range of 10^{-6} to 10^{-9} g adsorbed on the emitter). (2) Polar compounds of low volatility display high molecular ion intensities and little or no thermal fragmentation. (3) The high molecular ion intensities exhibited in fdms are of particular importance in metabolic studies since extracts of living cell material are very difficult to purify completely when only small quantities of the substances are present. In the case of fd mass spectra, the impurities also give only molecular ions, resulting in a considerably simplified spectrum. This enables easier identification of the metabolites and/or nonmetabolic decomposition products.

EXPERIMENTAL SECTION

The ei spectra were produced on a LKB 9000 (70 eV) instrument and all fd mass spectra were obtained with a CEC 21-110 B, the solvent for all compounds being acetone. High temperature activated emitters (Schulten and Beckey, 1972a) were used. The 10 μm tungsten wires adsorb enough of the sample to expose the photoplates for between 5 and 15 min. Ilford Q2 plates were used if not otherwise stated. The ion currents measured between the field anode and the slotted cathode plate were about 2×10^{-8} A. The spectra in Figure 7a,b were obtained by dipping the emitter into a cold (20°) saturated solution of the samples. The same emitter was used for both measurements, with the emitter heating current being 12 mA in both cases. All spectra of the aldrin-4,5-*cis*(*trans*)-diols (Figures 6a,b and 7a,b) were registered under comparable low resolution conditions.

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RESULTS AND DISCUSSION

Field Desorption Emitters. There has been rapid progress in the development of production of field anodes (Beckey, 1971) with high emission properties, mechanical, and chemical stability since the introduction of fdms of organic molecules (Beckey, 1969). On emitters activated with benzonitrile, at high temperatures microneedles grow that consist of carbon. They are resistant to corrosive chemical attack and enable fd measurements of perfluorinated alkanes, used for mass references (Schulten and Beckey, 1972a), to be made. They are also morphologically unchanged after repeated use for the fdms of chlorinated pesticides. Thus they are especially suitable for this latter class of compounds, since a large number of samples can be measured with one high temperature activated emitter without deterioration of the emission properties.

Pesticides. Figure 1a shows the eims of endrin. The $M \cdot^+$ peak (m/e 378) is unambiguously detectable, although only about 5% of the intensity of the base peak (m/e 263). The large number of peaks, in this case about 300, which is advantageous for the extraction of structural information, nevertheless can impede the detection of a pesticide or its metabolites in a contaminated sample.

In contrast, Figure 1b, the electrically recorded fd mass spectrum, contains no fragment in the mass range between m/e 300 and the molecular ion group. However, since the sample desorbs in a relatively small time interval from the fd emitter, the time available for scanning the magnet over the whole mass range is limited when using electric detection. For extremely polar compounds such as salts (*e.g.*, phosphates, sulfates, and acetates), the desorption time from the emitter is very critically dependent on the emitter temperature (Schulten and Beckey, 1972a) and may be only a few seconds. Moreover, the ion currents obtained are considerably smaller in fd compared to ei, so that further loss of intensity due to fast scanning becomes unacceptable. To overcome this difficulty one can use a computer in the time-averaging mode or, as is shown in the following examples, photographic recording, which is preferable for high resolution work (Schulten and Beckey, 1972b).

Figure 2 shows the fd mass spectrum of endrin recorded on a vacuum-evaporated AgBr photoplate (available from Ionomet Co., Burlington, Mass.). At room temperature, only the molecular ion group is detectable with no fragments, but raising the emitter temperature to about 200° over 5 min results in a thermally-induced fragmentation. High resolution mass measurements (20,000 resolution, 10% valley definition) show that the C_{12} cage of the molecule stays intact and that fragmentation is entirely due to consecutive loss of one or more Cl atoms and HCl groups, which is only the initial fragmentation step under ei conditions.

Using the same resolution, the fd mass spectrum of heptachlor (Figure 3) was obtained. However, in this case,

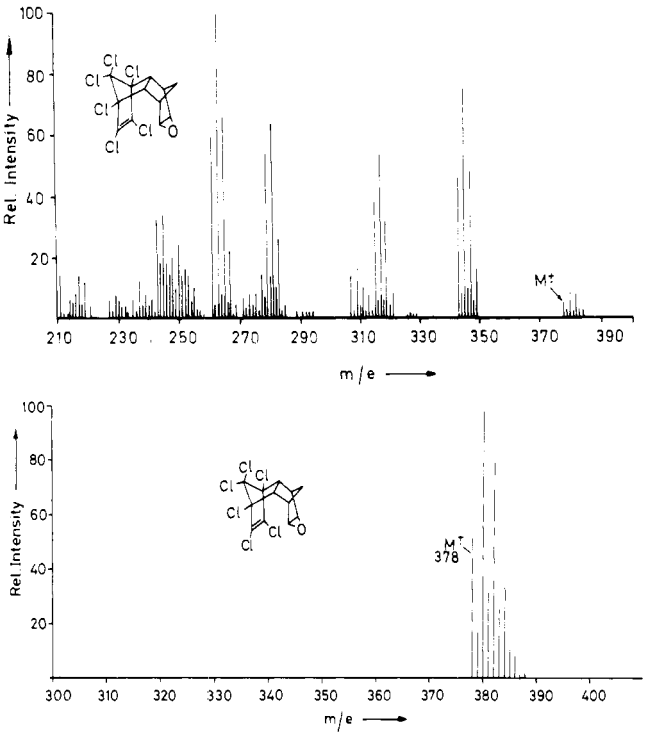

Figure 1. a. (top) Endrin eims; b. (bottom) Endrin fdms, electric recording.

the three fragments originate from C–C cleavages, when the emitter heating current is varied from 0 to 50 mA over 20 min (the 10 μm tungsten emitter is at red heat at the end of the exposure). The precise mass determination indicates a $C_2H^{35}Cl$ fragment at m/e 59.977 and two peaks at m/e 218.995 and 307.889. The probable elemental composition and origin of the fragments are given in Figure 3. It must be pointed out that with low resolution and electric recording in the mass range between m/e 200 and the molecular ion group, normally no fragment is detectable. The fast appearing thermal desorption products can, however, be recorded with an integrating detection system. The base peak in both cases is again in the molecular ion group. The simple spectrum contrasts with the eims (Damico *et al.*, 1968) which display some 60 peaks in the mass range from m/e 210 to m/e 380 alone, and a $M\cdot^+$ intensity of less than 10% of the base peak (m/e 100).

METABOLITES

It is a common observation that in a living organism a nonphysiological compound is degraded to a metabolite of higher polarity. The following examples show the advantages of using fdms as opposed to eims for the identification of such metabolites. One metabolite of isodrin, recently isolated from white cabbage leaves (*Brassica oleracea* var. capitata), is a carboxylic acid (Klein and Weisgerber, 1972). The comparison of the eims (Figure 4a) and the high resolution fdms (18,500 resolution, 10% valley definition) (Figure 4b) of this material (hexachloro-5-norbornene-2,3-dicarboxylic acid) clearly indicates the difference. There is no $M\cdot^+$ ion in the ei mass spectrum, while it is the base peak in fdms. The peak at the highest mass in the eims is m/e 368 ($M - H_2O$). The loss of water probably occurs by a thermal process prior to ionization. In fdms the acid moiety produces high $(M + 1)^+$ intensities due to proton transfer reactions in the adsorbed surface layer (Röllgen and Beckey, 1970, 1971). On the other hand, protonation of chlorine and thermal elimination lead to the two peaks of hydrochloric acid at m/e 35.977 and 37.974. The third fragment at m/e 45.006 (CO_2H) is often observed in fdms of carboxylic acids; *e.g.*, amino acids (Winkler and Beckey, 1972). Water elimination or other fragmentations are not observed under our ionization and detection conditions.

In attempting to obtain the maximum resolution with the CEC 21-110B instrument, the fdms of isodrin-4,5-*cis*-diol was examined (Figure 5). The resolution achieved is as good as that obtained in the ei and fi modes on the same instrument (Schulten and Beckey, 1972b). Taking into account the logarithmic blackening of the photoplate, the correct isotope ratios of the parent ion group for six Cl atoms are found. The precision of the mass determination is good. Mass measurements were made using perfluorotributylamine as the reference compound, with the average mass error being approximately 5 millimass units.

Using eims, one cannot differentiate between aldrin-4,5-*cis*- and aldrin-4,5-*trans*-diol, a known metabolite of dieldrin in rats (Korte and Arent, 1965), as is demonstrated by Figure 6a,b. The fdms of these compounds (Figure 7a,b) demonstrates a significant difference. Again the molecular ion is considerably enhanced and only two important fragments are detectable. One is the usual loss of chlorine, which is generally observed to be the first fragmentation step in eims. The other, due to a loss of water, is absent in eims, but is very prominent in the field desorption mass spectra. The process of water elimination

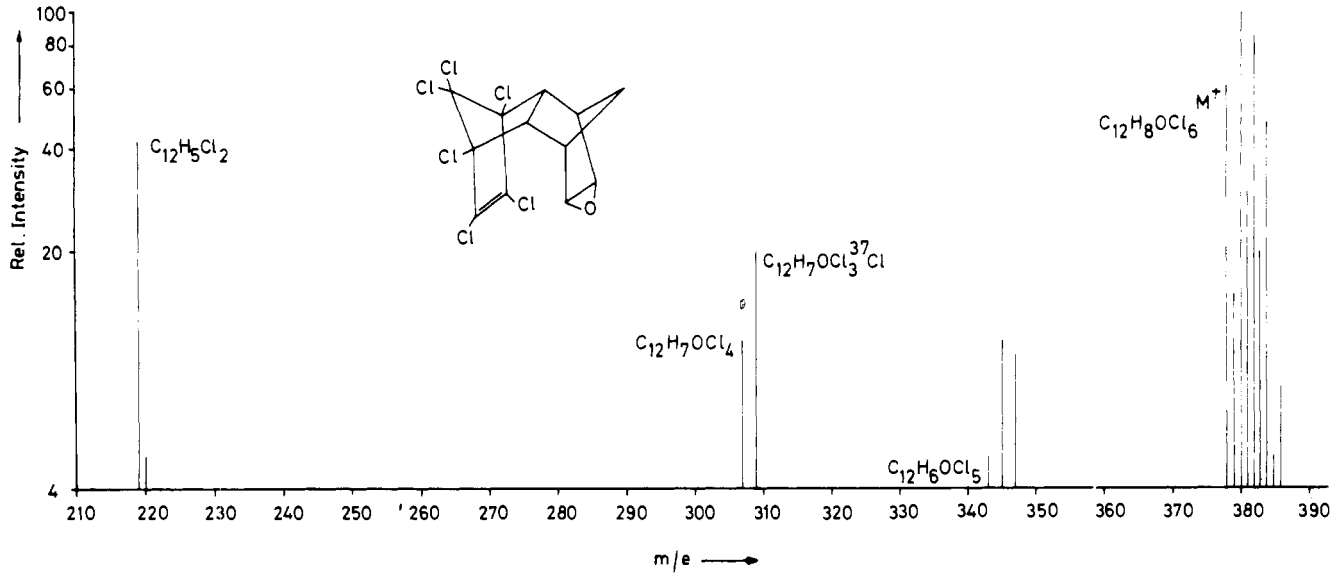

Figure 2. Endrin fdms, photographic recording; vacuum-evaporated AgBr plate, high resolution; emitter temperature, $\approx 200°$.

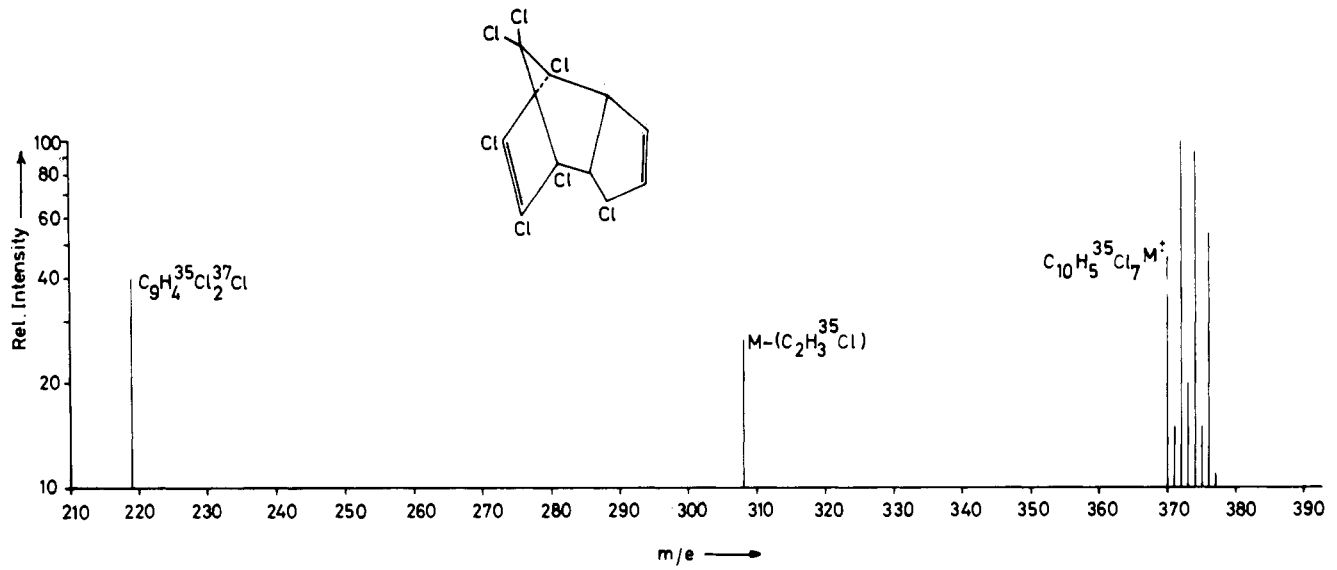

Figure 3. Heptachlor, fdms.

is dependent on the stereochemistry of the molecule. The trans isomer displays a peak at m/e 378 (M − H_2O) which is more than 20 times larger than the m/e 378 of the cis isomer. This is consistent with the usual chemical experience, but in the case of fdms, it is probable that the high field and the emitter surface also play an important role.

Further investigations of this phenomenon are currently being pursued.

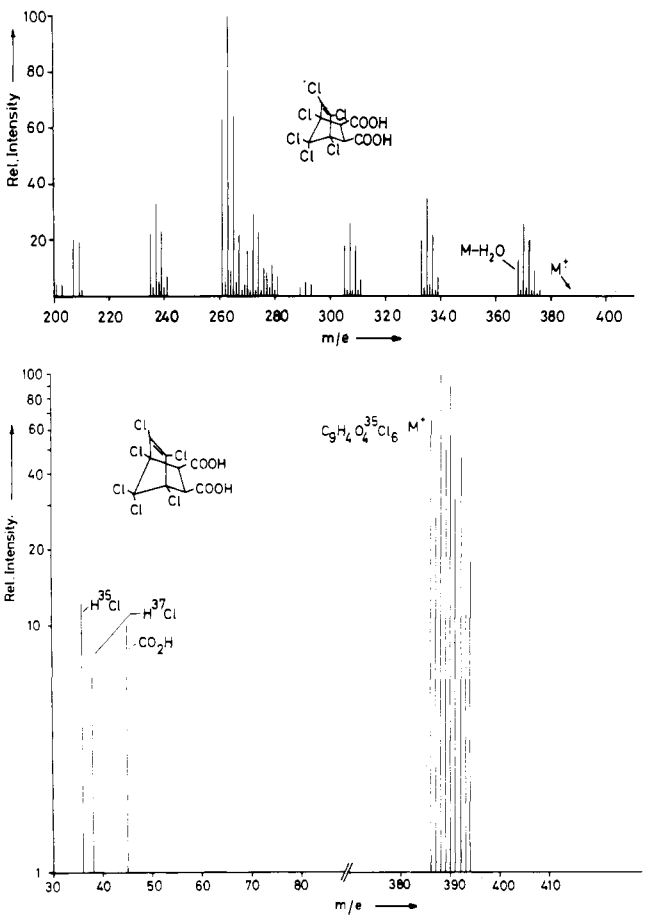

Figure 4. a. (top) 1,4,5,6,7,7-Hexachloro-5-norbornene-2,3-*endo*-dicarboxylic acid, eims; b. (bottom) 1,4,5,6,7,7-Hexachloro-5-norbornene - 2,3-*endo*-dicarboxylic acid, fdms.

CONCLUSION

To the best of the authors' knowledge, there was no analytical method available for positive identification of thermal highly unstable metabolites in the submicrogram range. Although recent investigations (Biros *et al.*, 1972; Dougherty *et al.*, 1972) show a remarkable potential for the detection of the basic polycyclic chlorinated pesticides with the chief emphasis on the commonly-used parent insecticides, with increasing polarity due to metabolism, however, the molecular or quasimolecular ions are significantly reduced with CI but remain the base peaks with fdms (Schulten *et al.*, 1972). Considering this point, field desorption mass spectrometry offers a solution to the problem especially relevant in environmental investigations. The use of fdms in studies of drug metabolism and toxicology has already been described (Schulten and Beckey, 1972c). The results presented here indicate their usefulness in both analysis of pesticides and drugs and their metabolites.

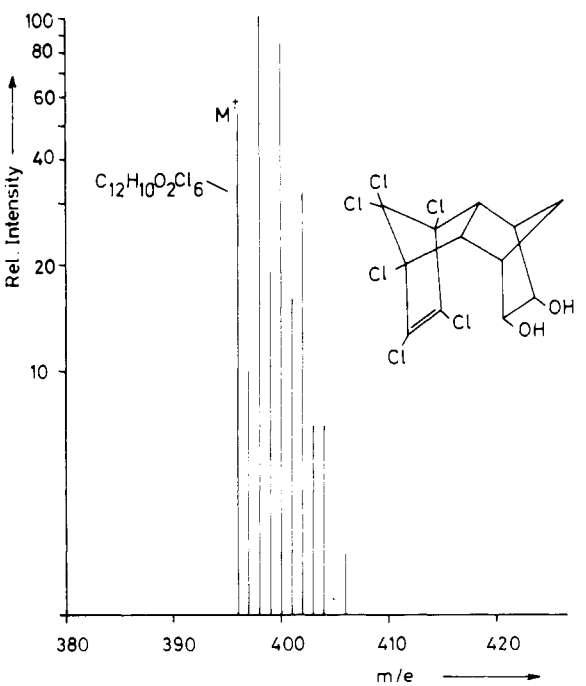

Figure 5. Isodrin-4,5-*cis*-diol, fdms; resolution 28,900 (10% valley definition).

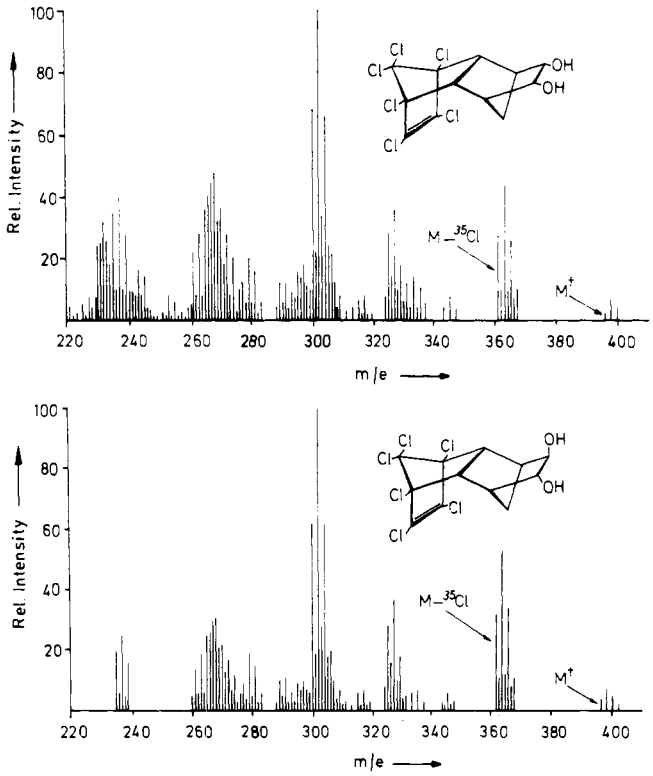

Figure 6. a. (top) Aldrin-4,5-*cis*-diol, eims; b. (bottom) Aldrin-4,5-*trans*-diol, eims.

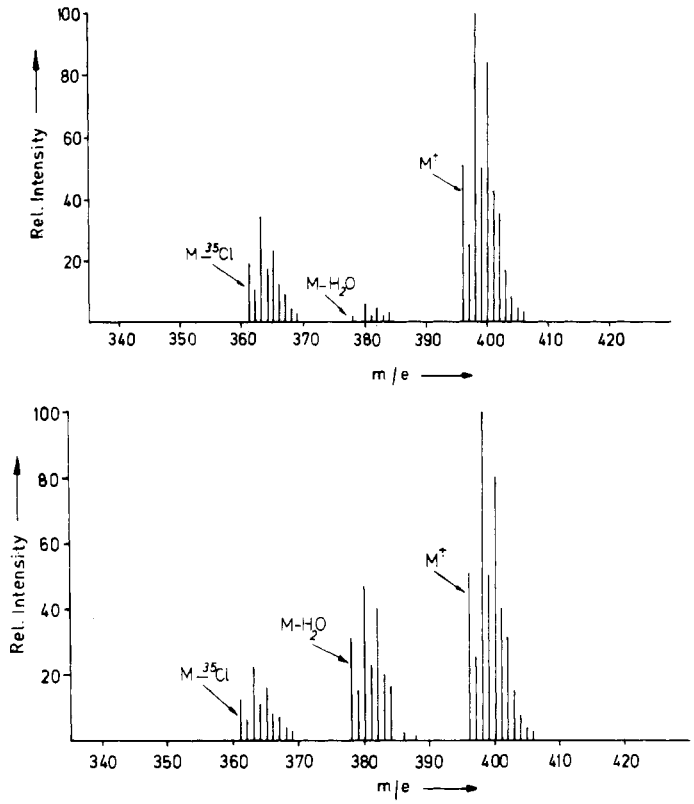

Figure 7. a. (top) Aldrin-4,5-*cis*-diol, fdms; b. (bottom) Aldrin-4,5-*trans*-diol, fdms.

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LITERATURE CITED

Beckey, H. D., "Field Ionization Mass Spectrometry," Pergamon Press, Oxford, and Akademie Verlag, Berlin, 1971.
Beckey, H. D., *Int. J. Mass Spectrom. Ion Phys.* **2,** 500 (1969).
Biros, F. J., Dougherty, R. C., Dalton, J., *Org. Mass Spectrom.* **6,** 1161 (1972).
Damico, J. N., Barron, R. P., Ruth, Y. M., *Org. Mass Spectrom.* **1,** 131 (1968).
Damico, J. N., Barron, R. P., Sphon, J. A., *Int. J. Mass Spectrom. Ion Phys.* **2,** 161 (1969).
Dougherty, R. C., Dalton, J., Biros, F. J., *Org. Mass Spectrom.* **6,** 1171 (1972).
Klein, W., Weisgerber, I., Institut für Oekologische Chemie, 5205 St. Augustin, West Germany, personal communication, 1972.
Korte, F., Arent, H., *Life Sci.* **4,** 2017 (1965).
Röllgen, F. W., Beckey, H. D., *Surface Sci.* **23,** 69 (1970).
Röllgen, F. W., Beckey, H. D., *Surface Sci.* **27,** 321 (1971).
Schulten, H. R., Beckey, H. D., *Org. Mass Spectrom.* **6,** 885 (1972a).
Schulten, H. R., Beckey, H. D., *Org. Mass Spectrom.* submitted for publication (1972b).
Schulten, H. R., Prinz, H. A., Beckey, H. D., Tomberg, W., Klein, W., Korte, F., *Chemosphere* submitted for publication (1972).
Schulten, H. R., Beckey, H. D., 20th Annual Conference on Mass Spectrometry and Allied Topics, Dallas, Texas, USA, (June 6, 1972c).
Winkler, H. U., Beckey, H. D., *Org. Mass Spectrom.* **6,** 655 (1972).

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